

Application No. 10/590,118
Paper Dated: February 6, 2009
In Reply to USPTO Correspondence of January 6, 2009
Attorney Docket No. 4544-062454

300-600 ml of chilled acetone vortexing it for 10-20 ~~min~~-minutes and filtering it through ~~whatman~~-Whatman No. 1. This step was repeated ~~till~~-until the lipids in the flask became whitish or colorless. This was filtered through ~~whatman~~-Whatman No. 1 and the filtrate was discarded. The lipids present on the filter paper were dissolved with C:M-chloroform:methanol (2:1) and transferred to the R-Bround bottom (RB) flask. The solvent ~~Solvent~~ was rotary evaporated under reduced pressure at 40-50° C. The crude preparation was reconstituted in 10-16 ml of C:M (2:1) and stored at -20° C. for further use. --

MD
1/12/10 Please replace the paragraphs beginning on page 3, line 23 and ending on page 4, line ¹³~~14~~, with the following rewritten paragraphs:

-- The Silica-silica gel H (S.D. Fine Chemical, India) was activated at 100-110° C. for 1-1.30 ~~hrs~~-hours (~~Hot~~-hot air oven) was packed with, glass column (2.6.times.30 cm) with manual tapping ~~and in which one end was plugged with a cork and a known quantity of crude material (4~~ g1.0 g/5 ml, stock) was loaded on either side of the column another side. The column was run in an ascending direction in a on chromatographic jar (4.5.times.25 cm) with 150-200 ml of purification solvent, ~~160-200 ml~~ (mobile phase) in a ratio of 66:25:4 (C:M:W)^{sup.7,8} chloroform:methanol:water at room temperature ~~to run the column till other it reached the end~~ following the procedure in reference 7.

The column was removed from the chromatographic jar and placed on fume hood to evaporate the solvent from the column. ~~The~~ A 10 cm length of each fraction was carefully scrapped using clean glass rod ~~so as to get the separate the individual molecules which that~~ were adsorbed with the silica gel depending upon the mobility and Retardation Factor (RF) value (46.6, 63.4, 68.3, 67.2 and 72.4%) of the individual molecul molecules. The individual ~~fraction~~-fractions ~~werewas~~ collected and placed into clean dry glass test tubes, which were labeled with respective fraction number, Ten ml of extraction solvent (mixture of chloroform: ~~Methanol~~-methanol 2:1) was added to each test tubes and kept at room temperature for 30-40 ~~min~~-minutes The purity of eluted material was analyzed by TLC and the selected fraction were further filtered through Whatman filter paper No. 1 to remove the silica gel from the samples. The pure fractions were pooled and these were characterized by ~~conventional methods (Immuno staining on TLC, ELISA and by~~